S.pneumoniae topo IV relaxation kits

Product Description (Product Numbers SPR4001 and SPR4002)

S.pneumoniae Topoisomerase IV is prepared by overexpressing the parC and parE subunits in *E. coli* and purifying them by in-house methods adapted from Peng and Marians, 1999. It is supplied as a heterotetramer complex. The enzyme is supplied at a minimum concentration of 5 -10 U/µl in Dilution Buffer. Kit supplied with enzyme, assay and dilution buffers and supercoiled plasmid substrate.
Store at -80 °C. (Stable for 6 months undiluted.)
It is recommended that the enzyme is aliquoted to avoid repeated freeze-thaw cycles.
For *in vitro* laboratory research use only.

Dilution Buffer

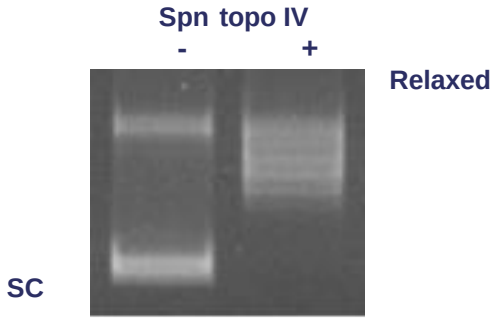
40 mM HEPES.KOH (pH 7.6)
100 mM potassium glutamate
1 mM DTT
1 mM EDTA
40 % (v/v) glycerol

Assay Buffer (supplied as 5X stock)

40 mM Tris-HCl (pH 7.5),
6 mM $MgCl_2$,
10 mM NaCl,
10 mM DTT,
200 mM potassium glutamate,
1 mM ATP,
0.05 mg/ml BSA

Relaxation Assay

1 U of *S.pneumoniae* topoisomerase IV will relax 0.5 ug of supercoiled plasmid DNA when incubated in 1X Assay Buffer in a total reaction volume of 30 µl at 37 °C for 30 minutes.
Gels are run in the absence of ethidium bromide or chloroquine.



Quality Control

Purity: The parC and parE subunits are purified to > 95 % purity as judged by SDS-polyacrylamide gel electrophoresis.

Reference

Peng, H. and Marians, K.J. (1999). Overexpression and purification of bacterial topoisomerase IV, in *DNA Topoisomerase Protocols* Vol. I (Bjornsti, M-A., and Osheroff, N. eds.), Humana Press, Totowa, N.Jersey pp.163-169